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First record on the biology of *Sarcophaga* (Bulbostyla) (Diptera, Sarcophagidae)

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Abstract

A first breeding record for *Sarcophaga* (*Bulbostyla*) *cadyi* Giroux & Wheeler on the American giant millipede *Narceus americanus* (de Beauvois) (Spirobolida, Spirobolidae) is reported. Digital photographs of the terminalia of *S.* (*B.*) *cadyi* and of *Sarcophaga* (*Bulbostyla*) *yorkii* Parker are also provided.

Keywords

feeding behaviour, flies, host, millipedes, Nearctic Region, Spirobolidae

Introduction

Sarcophaga Meigen is a large and diverse genus comprising about 890 valid species worldwide (Buenaventura et al. 2017). In Canada, 39 species are currently known including 21 species recorded in Quebec (Pape 1996; Dahlem and Naczi 2006; Giroux and Wheeler 2010). Adult flies of *Sarcophaga* feed on various resources including sugar, carrion and dung. The main resources of the larvae are dead arthropods, snails and small vertebrates which they can use as scavengers, parasitoids or predators (Pape 1996; Coupland and Barker 2004; Mello-Patiu 2016). Within this genus, the recently described subgenus *Bulbostyla* Giroux & Wheeler comprises nine species restricted to North and South America. It differs from other *Sarcophaga* mainly by characters of the

male genitalia (Giroux and Wheeler 2010). The ecology of *Bulbostyla* species remains little known, although most specimens were collected on hilltops (Giroux and Wheeler 2010). No feeding records have previously been documented for larvae.

Here, we present the first observation of an interaction between the flesh fly *S.* (*Bulbostyla*) *cadyi* Giroux & Wheeler and the American giant millipede *Narceus americanus* (de Beauvois) (Spirobolida, Spirobolidae). We also present digital photographs of the male terminalia of both species of *Bulbostyla* found in the province of Quebec (*S.* (*B.*) *cadyi* and *S.* (*B.*) *yorkii* Parker) and photographs of the female external terminalia of *S.* (*B.*) *cadyi*.

Materials and methods

A dead *N. americanus* colonized by eight sarcophagid larvae was collected on August 20, 2017. The millipede was found on the forest floor (45°33.18'N, 73°18.30'W) at Mont-Saint-Bruno National Park in southern Quebec. The millipede and the larvae of *S. cadyi* were brought to the laboratory and kept at constant room temperature (~20 °C) in a small plastic container with garden soil. Once adult flies emerged, they were killed in the freezer and preserved in 70% alcohol. In order to be morphologically identified, they were rinsed twice in 100% ethyl acetate, then dried and pinned.

The specimens of *S.* (*B.*) *yorkii* were collected using a hand-held entomological net at the summit of Mont Rigaud (45°27.96'N, 74°19.56'W, summers of 2007 and 2017) and of Mont-Saint-Bruno (45°33.12'N, 73°19.68'W, summer 2010). Those specimens were killed using ethyl acetate fumes and pinned shortly afterwards.

The habitus photographs (Figs 1–4) were taken using a Nikon D810 DSLR camera with Nikon Micro-Nikkor 200 mm f/4 lens on a Manfrotto 454 micrometric positioning sliding plate. Lighting was provided by two Nikon SB-25 flash units with a Cameron Digital diffusion photo box. Adobe Photoshop Elements 13 was used as post-processing software. Photographs of the terminalia and genitalia were taken with an Olympus DP27 camera mounted with stereoscope SZX16. Images were captured and stacked using Helicon Focus 7 before being enhanced using Adobe Photoshop CC (version 20.0) (Adobe Systems, Mountain View, CA).

To solidify species identity, a leg of some specimens of *S.* (*B.*) *cadyi* and *S.* (*B.*) *yorkii* were submitted to LifeScanner (http://lifescanner.net/) and others to the Canadian Centre for DNA Barcoding for DNA barcoding. It was only possible to obtain sequences for *S.* (*B.*) *cadyi*. Those sequences were compared and analysed using the Barcode of Life Data (BOLD: http://boldsystems.org/) System ID Engine (Ratnasingham and Hebert 2007). Individual sequences from the successful specimens are publicly available via GenBank accession codes MK585627–MK585630. They can also be retrieved from BOLD in the public dataset DS-SARCOPH (https://doi.org/10.5883/DS-SARCOPH).

The terminology of the terminalia follows Buenaventura and Pape (2017). All voucher specimens are deposited in Insectarium de Montréal's scientific collection (IMQC).

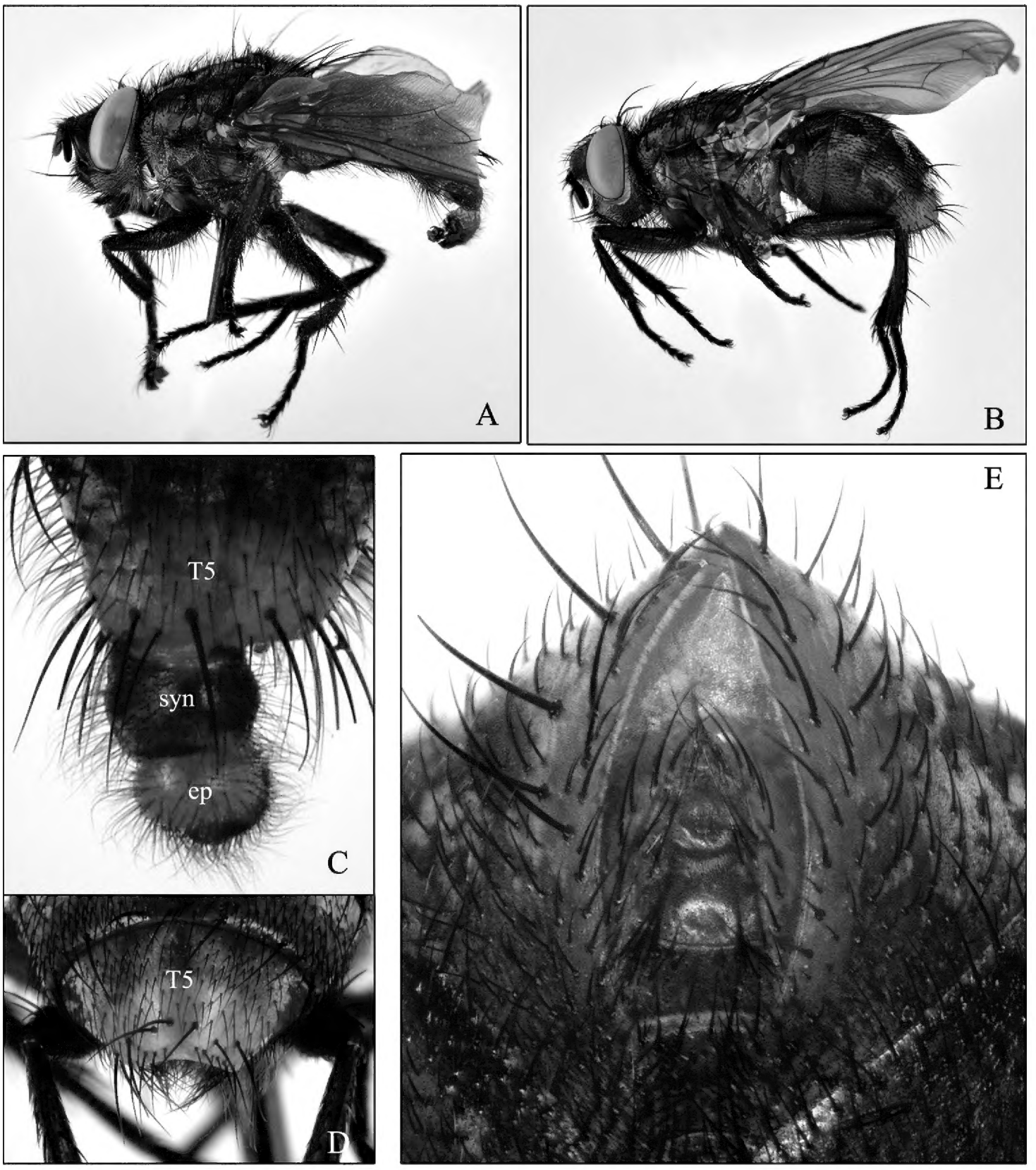

Figure 1. *Sarcophaga* (*Bulbostyla*) *cadyi* **A** male habitus **B** female habitus **C** male tergite 5 (T5), syntergosternite 7+8 (syn) and epandrium (ep), dorsal **D** female tergite 5 (T5), dorsal **E** female postabdomen, ventral.

Results and discussion

We present the first breeding record for a species of *Bulbostyla* and the first mention of their larvae developing in a spirobolid millipede. We also present the first mention of a *Sarcophaga* species showing a feeding interaction with a millipede in North America, and the second worldwide after the European species *Sarcophaga* (*Myorhina*) *iulicida* Pape (Pape 1990a).

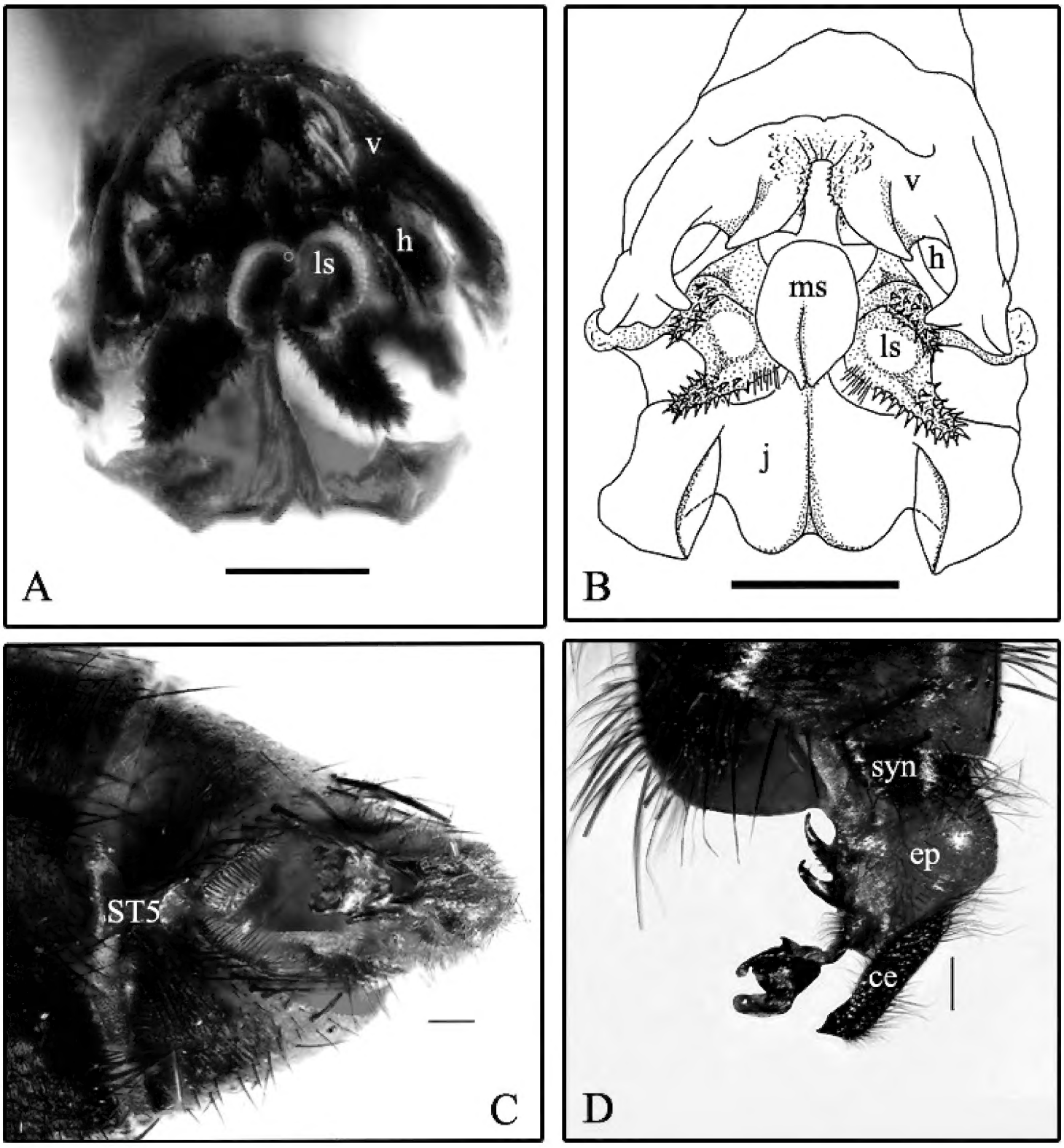

Figure 2. *Sarcophaga* (*Bulbostyla*) *cadyi* **A** distiphallus, anterior **B** distiphallus, anterior (from Giroux and Wheeler 2010) **C** male postabdomen, ventral **D** male postabdomen, left lateral. Abbreviations: j, juxta; ls, lateral stylus; ms, median stylus; h, harpes; v, vesica; syn, syntergosternite 7+8; ep, epandrium; ce, cercus; ST5, sternite 5. Scale bars: 0.2 mm (**A–B**), 0.5 mm (**C–D**).

Only three dipteran families (Sarcophagidae, Phoridae, Sciomyzidae) have been reported as parasitoids of diplopods (Hash et al. 2017). Within the Sarcophagidae, the species *Blaesoxipha beameri* Hall (Pape 1994) and species of the genus *Spirobolomyia* Townsend have been bred exclusively from spirobolid millipedes (Aldrich 1916; Pape 1990b, 1996) in North America. However, it is likely that species of this genus are not true millipede parasitoids. All observations of larviposition by *Spirobolomyia* species were on injured hosts with wounds large enough for the larva to enter (Hash et al. 2017).

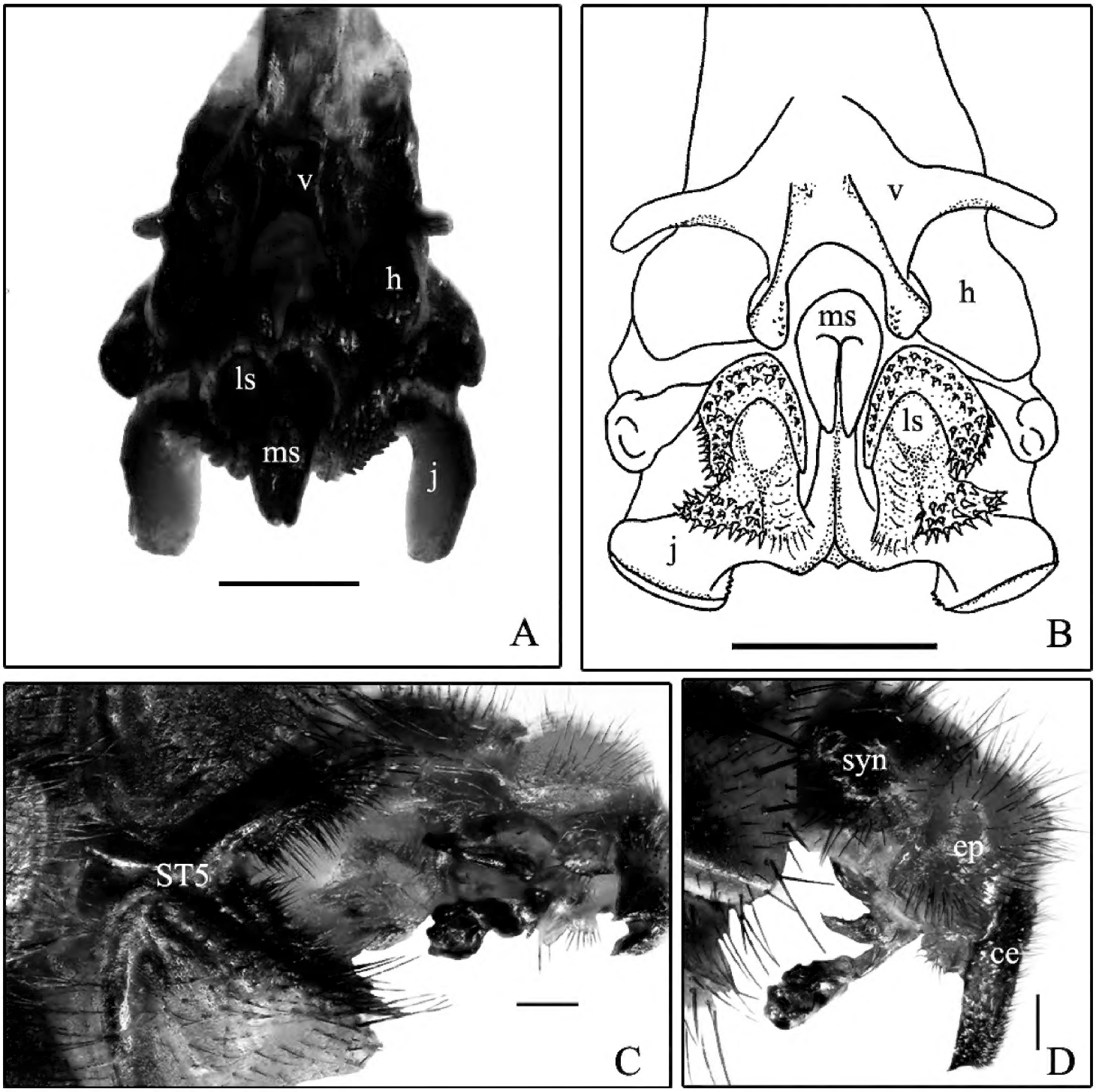

Figure 3. *Sarcophaga* (*Bulbostyla*) *yorkii* **A** distiphallus, anterior **B** distiphallus, anterior (from Giroux and Wheeler 2010) **C** male postabdomen, ventral **D** male postabdomen, left lateral. Abbreviations: j, juxta; ls, lateral stylus; ms, median stylus; h, harpes; v, vesica; syn, syntergosternite 7+8; ep, epandrium; ce, cercus; ST5, sternite 5. Scale bars: 0.2 mm (**A–B**), 0.5 mm (**C–D**).

We did not observe the larviposition of *S.* (*B.*) *cadyi* on *N. americanus*, which was already dead and colonized by the last instar larvae when we found it. Thus, we do not know if the spirobolid millipede was healthy, injured or already dead upon arrival of the female sarcophagid fly. In this sense, further investigations are needed to be able to determine the larval feeding habits of *S.* (*B.*) *cadyi*. The larvae pupated around August 25th. They pupated inside the millipede rather than exiting and pupating in the surrounding soil. It is unclear if this behaviour was due to laboratory conditions, or if it is also displayed in nature. Four males and four females emerged two weeks later, between 7 and 11 September 2017.

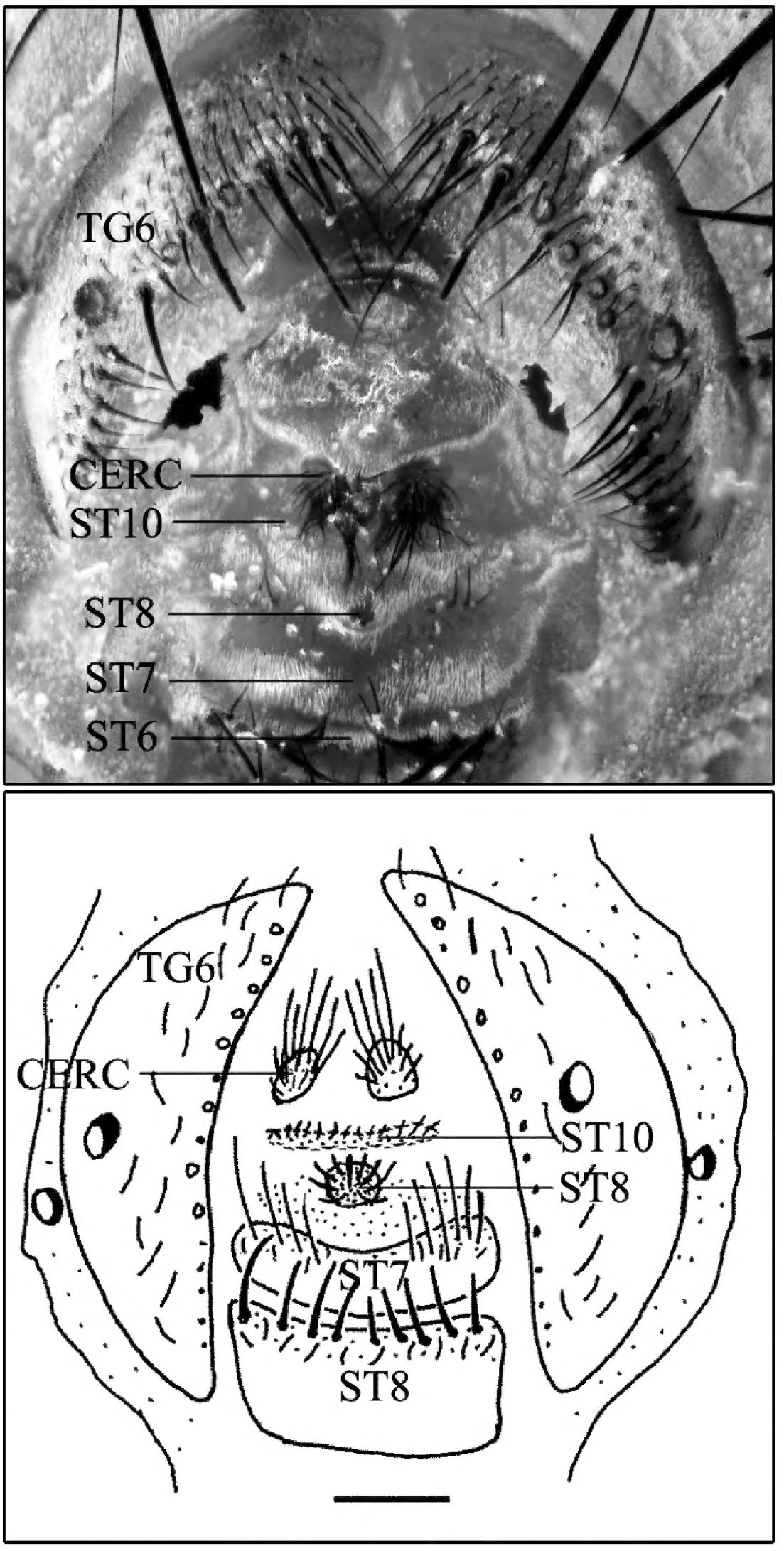

Figure 4. *Sarcophaga* (*Bulbostyla*) *cadyi* **A** female external terminalia, dorsoventral **B** female external terminalia, dorsoventral (from Giroux and Wheeler 2010). Abbreviations: cerc, cerci; st, sternite; tg, tergite. Scale bars: 0.5 mm (**A–B**).

Descriptions and an identification key for males of *S.* (*B.*) *cadyi* and *S.* (*B.*) *yorkii* can be found in Giroux and Wheeler (2010). However, in order to help in the identification of these species, some digital photographs are provided here: the habitus of *S.* (*B.*) *cadyi*, male and female (Fig. 1A, B); the postabdomen of a *S.* (*B.*) *cadyi* male (Figs 1C, 2C, D) and female (Fig. 1E) as well as the one of a *S.* (*B.*) *yorkii* male (Fig. 3C, D). Male and female specimens of both species have tergite 5 with an orange-yellow posterior half or third (sometimes entirely yellow) and a row of strong setae forming a semi circle that spreads on the apical third (Fig. 1C–E). The male cerci and syntergosternite 7+8 are darker than the epandrium (Figs 1C, 2D, 3D). The window on male sternite 5 is almost even with the rest of the base (Figs 2C, 3C). The male distiphallus of both species as well as the external terminalia of female *S.* (*B.*) *cadyi* were digitally photographed (Figs 2A, 3A, 4A) and illustrations (Figs 2B, 3B, 4B) were added for a better understanding of their structural morphology.

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